Corn fiber gum: A potential gum arabic replacer for beverage flavor emulsification [☆]

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Abstract

The US food industry would benefit from a domestically produced gum with a dependable supply and consistent quality that can be used for preparing oil-in-water (O-in-W) emulsions, such as citrus oil emulsions for beverages. Corn fiber gum (CFG) is an arabinoxylan (hemicellulose) extracted from the corn kernel pericarp and/or endosperm fiber fractions that can possibly fulfill this need. In this study two different types of CFG, CFG-1 and 2, were prepared from corn fiber collected from different wet or dry corn milling facilities by (a) sequential alkaline extraction and alkaline hydrogen peroxide bleaching and (b) an additional alkaline hydrogen peroxide treatment of the alkali treated residue, respectively. CFG-1 might be linked to the cell wall matrix through ester linkages and CFG-2 by non-ester linkages and/or other strong interactions. The stabilization of O-in-W emulsions by corn fiber and acacia gums was investigated by preparing emulsions with a high pressure homogenizer and monitoring the emulsion breakage by turbidity measurements. All CFG samples were effective emulsifiers but CFG-2 extracted separately from three different corn fiber sources was determined to be a better emulsifier than the corresponding CFG-1. CFGs isolated from wet milled pericarp and endosperm fiber and wet milled pericarp fiber have a higher protein content than CFGs isolated from dry milled pericarp fiber and were determined to be better emulsifiers for the O-in-W emulsion system. The emulsifying properties of all CFGs including an industrial grade commercial CFG were determined to be better than native and modified acacia gums.
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1. Introduction

Corn fiber gum (CFG) is an alkaline extract of corn "fiber", which is the main low value by-product of the corn wet and/or dry milling process. Corn fiber from these sources is primarily composed of cell-wall non-starch polysaccharides derived from corn kernel pericarp and/or endosperm tissues. CFG is an arabinoxylan (hemicellulose B) with unique high solubility and low viscosity. Corn fiber obtained from the wet milling industry, commonly known as "white fiber" is a mixture of coarse and fine fibers.

The coarse fiber originates from the kernel pericarp or hull and the fine fiber is the inner cellular fiber from seed endosperm (Singh, Doner, Johnston, Hicks, & Eckhoff, 2000). Corn bran is a by-product from the commercial corn dry milling process and can also be called corn pericarp fiber as it originates only from this portion of kernel. The hemicelluloses are poorly defined and have long been neglected from an applications point of view, perhaps due to being low-viscosity gums, a property not seen as desirable by many food industries. However, it is now recognized that there is a need for gums with improved secondary characteristics that allow them to be used as bulking, bodying, emulsion stabilizing, and protective colloid agents, i.e., in applications in which low-viscosity, high-solid gum solutions are required. It has been suggested that if CFG could be produced commercially, it could be used as an adhesive, thickener and stabilizer

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(Wolf, MacMasters, Cannon, Rosewell, & Rist, 1953) and as a film former and emulsifier (Whistler, 1993; Woo, 2001). However, despite repeated attempts by numerous researchers and corn millers to develop a commercial product, none of the past attempts led to a suitable quality product at an affordable price. Further, the detailed evaluation of the viscosity and emulsification properties of pure CFG has not been accomplished. Part of the purposes of this report is to characterize these properties. Gum arabic, a natural exudate of acacia trees, is widely used in the food industry. It has a unique combination of excellent emulsifying properties and low solution viscosity despite its high molecular weight. So far this gum is considered to be the best gum in a dilute oil-in-water (O-in-W) emulsion system (Garti, 1999). However, almost all the world's gum arabic supply comes from three production areas in the Sahelian region of Africa. About 75% is produced by the Republic of the Sudan; most of the remainder comes from Senegal, Mauritania, and Nigeria. It is produced by trees which take at least 6 years to establish and which grow in a belt just below the Sahara desert (Glicksman, 1983). The exudate masses are picked by hand. Its supply is variable, uncertain and subject to climatic, economic and political conditions in the region. World demand for gum arabic is increasing and import into the US alone has increased from 6250 to 7500 metric tons from 1989 to 1994 (National Trade Data Bank, Department of Commerce). Thus the availability of an effective domestic substitute for gum arabic would be desirable for US producers of citrus O-in-W beverage emulsions and stable flavor powders. In addition, creation of a high-valued gum from a low-valued corn milling by-product could benefit US corn growers and processors.

CFG has been reported to be a homogeneous, predominantly carbohydrate molecule (Whistler & BeMiller, 1956; Montgomery & Smith, 1957), with the following sugar composition reported by various groups: D-xylose (48–54%), L-arabinose (33–35%), galactose (7–11%), and glucuronic acid (3–6%), (Doner, Chau, Fishman, & Hicks, 1998; Hespell, 1998; Suguwara, Suzuki, Totsuka, Takeuchi, & Ueki, 1994; Saulnier, Marot, Chanliaud, & Thibault, 1995a; Whistler & BeMiller, 1956). The gum structure is highly branched with a β-(1–4)-xylopyranose backbone and α-L-arabinofuranose residues as side chains on both primary and secondary hydroxyl groups (Saulnier et al., 1995a). Most of the D-glucuronic acid residues are linked to the O-2 position of xylose residues of the main xylan backbone (Montgomery & Smith, 1957). Galactose and some xylose residues are attached to the arabinofuranosyl branches (Whistler & Corbett, 1955).

CFG, like many hemicelluloses, is not extractable with water but can be extracted with alkali and alkaline H_2O_2 from plant cell walls and so it is believed that it is cross linked with other cell wall components. Covalent linkages, ionic and hydrogen bonding between the cell wall components have been claimed by Carpita and Gibeaut

(1993). The possible linkages between hemicellulose and the cell wall components are mentioned in detail by Doner and Hicks (1997) explaining that ferulic, diferulic and p-coumaric acids play a very important role in cross linking them together (Saulnier, Vigouroux, & Thibault, 1995b). It is also clear that polyphenolics such as lignin can form alkali resistant linkages (ether linkages) with hemicelluloses, which can be cleaved by alkaline hydrogen peroxide treatment. In the cell wall, arabinoxylan and protein are also closely associated as supported by the finding of a stable linkage between hemicelluloses and protein in corn bran (Saulnier et al., 1995a) and rye bran (Ebringerova, Hromadkova, & Berth, 1994). The present investigation is carried out to study the emulsification properties of CFG from different corn milling fractions and to relate their structural characteristics with their ability to stabilize emulsions. In addition, the effect of extraction conditions on the emulsion properties of CFG was also studied.

2. Materials and methods

2.1. Materials

Corn fiber samples were kindly provided by ADM Research, Bunge (North America, Bunge Milling, Inc., St. Louis, MO) and Cargill Central Research (Minneapolis, MN). They were oven dried by the suppliers before shipping. Fiber samples were ground to a 20-mesh particle size using a Wiley mill and extracted with hexane to remove oil (Moreau, Powell, & Hicks, 1996). Starch was removed from the 20-mesh de-oiled fiber by treating with Termamyl α-amylase (a gift from Novo Nordisk Bioindustrials, Inc., Danbury, CT) (Doner et al., 1998; Doner & Johnston, 2001). Native acacia gum (NAG) and modified acacia gum (MAG) were obtained from TIC Gums, Inc. (Belcamp, MD). Commercially available industrial grade CFG (CFG-IG) was a gift from National Starch and Chemical Company, Bridgewater, NJ. Cold-pressed Valencia orange oil was supplied by Citrus and Allied Essences Ltd., Belcamp, MD. The Polytron homogenizer (Model PT 10/35) was purchased from Brinkmann, Westbury, NY and the EmulsiFlex-B3 high-pressure homogenizer was purchased from Avestin Inc. (Ottawa, Canada).

2.2. Standard proximate analyses

Moisture, protein ($N \times 6.25$) and ash contents of corn fiber and acacia gums were determined according to Approved Methods 44–19, 46–30 and 08–01 (American Association of Cereal Chemists, 1995), respectively.

2.3. Isolation of CFG

CFGs were isolated from de-oiled and de-starched corn fiber according to the alkaline hydrogen peroxide procedures of Doner et al. (1998); Doner and Johnston (2001) with some modification (Fig. 1). De-oiled and de-starched

corn fiber (50 g) was mechanically stirred into water (0.5 l) and NaOH (2 g) and Ca(OH)$_2$ (1.9 g) were added to make 2 meq alkali/g fiber in the extraction solution. The mixture was boiled with efficient mechanical stirring for 1 h. The reaction mixture was centrifuged at 6000g for 20 min and the supernatant was separated from the residue by decantation. The residue was resuspended into 400 ml water and boiled for 5 min with mechanical stirring. The reaction mixture was centrifuged as above and the supernatant was decanted and combined with the first supernatant. The residue was saved for further isolation of CFG and a cellulose/arabinoxylan mixture (CAX). To the total volume (0.8 l, pH 11.30) of the combined extract, 5.1 g H$_2$O$_2$ (17 ml, 30%) was added which dropped the pH to about 9.60. The pH was readjusted to 11.5 by adding 50% NaOH ($\approx$ 17 ml) and stirred at room temperature for 2 h.

The pH of alkaline H$_2$O$_2$ extract was then adjusted to 4.0–4.5 by adding Conc. HCl ($\approx$ 17 ml) to precipitate Hemicellulose A (acid-insoluble arabinoxylan, "Hemi. A"), which was collected by centrifugation at 10,000 g for 30 min. Two volumes of ethanol (1.6 l) were gradually added to the supernatant (0.8 l) with stirring to precipitate the major arabinoxylan fraction, Hemicellulose B, or Hemi. B, (CFG-1). The CFG-1 was allowed to settle out as a white flocculent precipitate at the bottom of the beaker for 10–15 min. The clear alcohol/water mixture above the precipitate was removed by decantation. The white flocculent precipitate was transferred into another beaker, stirred in 100% ethanol and filtered under vacuum. The white residue obtained on the Buchner funnel was washed with 100% ethanol and dried in a vacuum oven at 50 °C overnight.

The residue left after alkali extraction was further extracted with alkaline H$_2$O$_2$ (0.1 g H$_2$O$_2$/g fiber, pH 11.5) at a boiling temperature for 1.5 h. The residue was removed by centrifugation (6000g, 15 min) and the pH of the supernatant was adjusted to 4.0–4.5 to precipitate Hemi. A. The precipitate was then removed by centrifugation at 10,000g for 30 min. The CFG-2 was obtained from the supernatant by precipitating it with 2 volumes of ethanol as a white flocculent precipitate, collected and dried as above.

2.4. Determination of carbohydrate composition

Sugars were analyzed by HPAEC-PAD using methanolysis (Chambers & Clamp, 1971; Svetek, Yadav, & Nothnagel, 1999) combined with TFA hydrolysis. The gum samples to be analyzed were first dissolved in de-ionized water (1 mg/ml). An aliquot of 100 µl of this solution along with 100 nmoles myo-inositol (internal standard) were dried in a Teflon-lined screw cap glass vial by blowing with filtered nitrogen followed by drying in a vacuum oven at 50 °C overnight. These samples were methanolyzed with 1.5 M methanolic HCl in the presence of 20% (v/v) methyl acetate for 16 h, cooled to room temperature and dried by blowing with filtered N$_2$ after

adding five drops of t-butanol. The methanolyzed samples were hydrolyzed with 0.5 ml 2 M TFA at 121 °C for 1 h, evaporated by blowing with filtered N$_2$ at 50 °C and the residue was washed by sequential addition and evaporation of three aliquots (0.5 ml) of methanol. In three separate glass vials were placed 100, 300 and 500 nmoles of a mixture of standard sugars containing fucose, arabinose, rhamnose, galactose, glucose and xylose. Then, 100 nmoles of myo-inositol (internal standard) was added to each vial, evaporated and dried as above. These standard samples were also methanolyzed and hydrolyzed as described above and used for quantification.

Hydrolysates were analyzed for neutral monosaccharide content by HPAEC-PAD using a Dionex ICS-2500 system that included a CarboPac PA20 column and guard column, a EG 50 eluent generator that produced the isocratic 10 mM KOH mobile phase, a CR-ATC continuously regenerated anion trap column, a GP 50 pump operated at 0.5 ml/min, an ED50 electrochemical detector utilizing the quadruple potential waveform, and an AS50 auto-sampler with a thermal compartment (30 EC column-heater). Acidic monosaccharide content was determined with a Dionex DX-500 system, which included a GP50 gradient pump, a CarboPac PA20 column and guard column, an ED40 electrochemical detector (gold working electrode and pH reference electrode), an LC25 chromatography oven (30 °C), a PC10 pneumatic controller (post-column addition of 500 mM NaOH), and an AS3500 autosampler. The mobile phase consisted of isocratic 10 mM CH$_3$COONa and 1 mM NaOH eluant for 10 min and then a linear gradient of 100–130 mM CH$_3$COONa in 100 mM NaOH for the following 20 min.

2.5. Emulsion preparation

Stock gum solution: Solutions of 6% gum with 0.12% preservative (sodium benzoate) and 0.36% citric acid were prepared by slowly adding the gum sample with vigorous stirring to a solution of sodium benzoate and citric acid in water at ambient temperature and then gently stirring overnight to produce a homogeneously hydrated solution (Yadav & Nothnagel, 2006). An adjustment for moisture content of the powders was done during the solution preparation.

Emulsion concentrate: The O-in-W emulsion concentrates were prepared in triplicate with the following formula: Valencia orange oil 10.0% (w/w); gum sample 5.0% (w/w); sodium benzoate 0.1% (w/w); citric acid 0.3% (w/w) and water to make up to 100% (w/w). To obtain this formulation, 2.5 g of the stock gum solution was taken into a 4 ml glass vial, and then 300 mg (360 µl) of orange oil and sufficient de-ionized water (200 µl) were added to make 3.0 g total solution. No weighting agent was used. The two phases of the solution were premixed by vortexing for a few minutes. Then the coarse emulsion was prepared by pre-homogenization using a polytron bench top homogenizer equipped with a 12 mm diameter head (Brinkmann, Switzer-

land, PT 10/35) at 15,000 rpm for 30 s. A fine emulsion was then prepared by passing the coarse emulsion through the EmulsiFlex-B3 high-pressure homogenizer (Avestin Inc., Canada) operating with 340 kPa (50 psi) regulator pressure which, after hydraulic amplification, corresponds to 68 MPa (10,000 psi) homogenization pressure.

Diluted emulsion: The resulting homogenized concentrates were diluted 125 × to 375 ml in a 10.0% (w/w) sucrose solution containing 0.1% (w/w) sodium benzoate and 0.3% (w/w) citric acid. Both the polytron homogenizer probe and high pressure homogenizer chamber were rinsed with three aliquots (3 ml each) of the sugar solution (saved from the total 375 ml solution for this purpose). Each time the probe was dipped in the solution and run for a few seconds, and then the same solution was passed through the high pressure homogenizer sample chamber. After these three rinses the high pressure homogenizer was rinsed with one passage of air to blow the remaining traces of liquid out of the sample chamber. All three rinses were combined in the 375 ml total solution in a 500 ml glass bottle.

2.6. Determination of emulsifying properties

The emulsion stability (ES) of the diluted emulsions was determined as turbidity (Pearce & Kinsella, 1978) with modification as explained by Einhorn-Stoll, Weiss, and Kunzek (2002). $T = 2.303AD/l$, where T = turbidity in 1/cm, A = observed absorbance at 650 nm, D = dilution factor, and l = path length of the cuvette in cm. A higher turbidity is an indication of greater ES. The absorbance of the diluted emulsion was measured immediately in a 1-cm path length cuvette at a wave length of 650 nm using a UV-Visible 160 Spectrophotometer (Shimadzu, Columbia, MA) to determine EA as turbidity. The bottles containing the emulsion were set on the bench at ambient temperature (22–24 °C) or in an oven at 45 °C without agitation for

gravity separation. The emulsion breakage was monitored by absorbance (loss of turbidity) measurement at 650 nm daily to determine the ES. The ES study was done for 14 days (for the experiment conducted at room temperature) or 7 days (for the experiment conducted at 45 °C). A 10.0% sugar solution containing 0.1% sodium benzoate and 0.3% citric acid was used as a blank for the absorbance measurements. The bottles containing emulsions were photographed to compare, document and record their emulsion breakage pattern (not shown).

3. Results and discussion

3.1. Isolation of CFG

CFG-1 and 2 were extracted from wet milled pericarp fiber (WPF), dry milled pericarp fiber (DPF); and wet milled pericarp and endosperm fiber (WPEF) according to the scheme shown in Fig. 1. This extraction procedure is a slight modification of the published isolation method for CFG (Doner et al., 1998; Doner & Johnston, 2001). Corn fiber produced with any milling procedure contains some percentage of oil which was removed by hexane extraction (Moreau et al., 1996). A considerable amount of starch is usually associated with fiber fractions, which was removed by using Termamyl α-amylase before CFG extraction. The CFG-1 was extracted from the destarched CF sample by a mixture of NaOH (0.1 M) and Ca (OH)$_2$, (0.05 M), containing 1 meq of each alkali/gram of fiber in the reaction medium at the boiling temperature and treating the alkaline extract with 30% H$_2$O$_2$ at pH 11.5 to bleach the product (Doner et al., 1998). This treatment released most of the CFG-1 linked through phenolic or non-phenolic ester linkages or cross-linked by diferulic ester bridge (Chanliaud, Saulnier, & Thibault, 1994). The amount of CFG-1 released by this treatment was about 22–26% of the total weight of the de-oiled and starch free
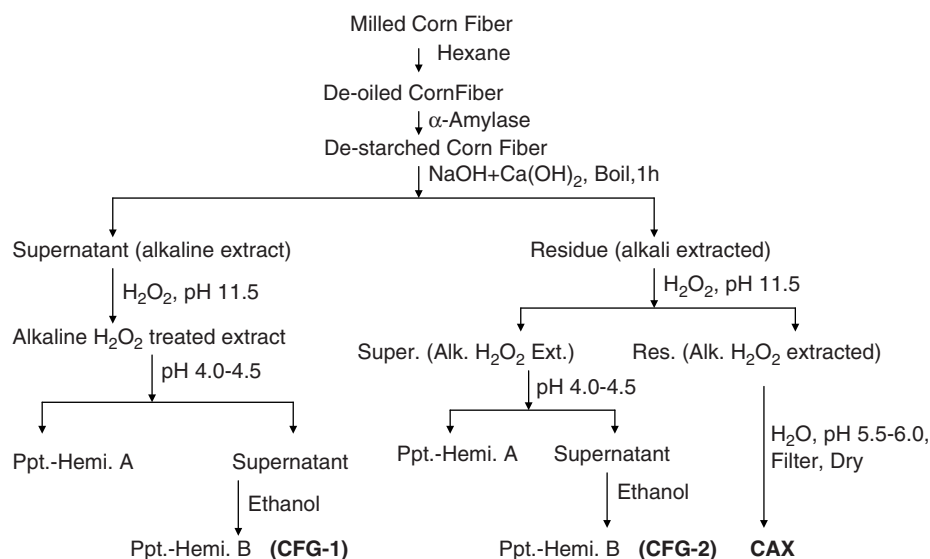

Fig. 1. Basic scheme for the isolation of corn fiber gum from corn fiber.

Table 1
Percentage yields[a] of corn fiber constituents

Samples	CFG-1	CFG-2	Hemi. A	CAX	Total yield
Wet milling pericarp fiber (WPF)	25.8	13.8	1.6	33.2	74.4
Dry milling pericarp fiber (DPF)	26.6	23.6	1.0	29.8	81.0
Wet milling pericarp and endosperm fiber (WPEF)	22.2	12.0	1.0	27.2	62.4

[a]Based on de-oiled and de-starched corn fiber weight.

fiber (Table 1). The alkali treated residue was further extracted with alkaline H_2O_2 (0.1 g H_2O_2/g fiber, pH 11.5) at boiling temperature to release additional arabinoxylans (CFG-2) held in the cell wall through more alkali resistant linkages, such as ether linkages. Polyphenolics like lignin can form alkali resistant ether linkages with arabinoxylans (Fincher and Stone, 1986; Hatfield, 1991; Jung and Deetz, 1991; Morrison, 1974; Watanabe, Ohnishi, Yamasaki, Kaizu, & Koshijima, 1989) and arabinoxylans can also be linked with the cell wall matrix by other alkali resistant linkages. CFG-2 could also be bound to the cell wall matrix by strong alkali resistant ionic and hydrogen bonding or by hydrophobic interactions. Protein connected to hemicelluloses by linkages that are stable to alkali and/or H_2O_2 may be present on both CFG-1 and 2. Their actual linkages are not known. They could be linked to each other by covalent bonds or the protein might be co-precipitated with arabinoxylans. Stable linkages between protein and arabinoxylans have been reported in corn bran (Saulnier et al., 1995a) and rye bran (Ebringerova et al., 1994). These proteins could be related to maize hydroxyproline-rich glycoprotein HRGP (as reported by Hood, Shen, & Varner, 1988; Hood, Hood, & Fritz, 1991) in maize pericarp. Glycosidic bonds to hydroxyproline residues in protein are known to be stable to alkali. Corn endosperm is known to contain zein proteins with hydrophobic amino acid residues. Proteins of this type could also impart emulsifying activity to a polysaccharide to which it was covalently attached.

The yield of CFG-2 varies from 12–24% of the total weight of de-oiled and starch free fiber (Table 1). The yield of CAX which is not characterized in this study but reported in Table 1 varies from 27 to 33% of the oil and starch free fiber.

3.2. Proximate analysis

The proximate analysis of corn fiber and acacia gums is given in Table 2. The moisture content of these gum samples (5.94–11.04%) was high enough to justify adjustment for the moisture in emulsion preparation. The ash content ranged from 2.7% to 16.97%, and industrial grade CFG had the highest amount of ash. The protein content was comparatively higher in CFG-2 than CFG-1 from each corn fiber fraction. The CFG-1 and 2 from wet milling had a higher protein content than that from dry milling fiber fractions. The commercial industrial grade CFG sample

Table 2
Proximate composition of corn fiber and acacia gums (wt%)

Samples	Moisture	Ash	Protein[a]
Wet milling pericarp fiber (WPF)			
CFG-1	8.41	2.70	1.90
CFG-2	7.68	5.20	2.88
Dry milling pericarp fiber (DPF)			
CFG-1	6.04	2.90	0.44
CFG-2	5.94	6.14	0.94
Wet milling pericarp and endosperm fiber (WPEF)			
CFG-1	6.06	3.22	1.31
CFG-2	6.87	4.83	3.75
Commercial samples			
CFG-IG (Industrial grade)	10.73	16.97	5.25
NAG (Native acacia gum)	8.43	3.91	0.44
MAG (Modified acacia gum)	11.04	3.3	1.75

[a]Calculated by multiplying nitrogen content by 6.25.

had the highest protein content (5.25%) among all the gum samples. The protein level of NAG was low but that of MAG fell within the ranges reported for the different species of acacia (Buffo, Reineccius, & Oehlert, 2001).

3.3. Sugar composition

Table 3 shows the sugar composition of corn fiber and acacia gums. No significant difference was observed in the general sugar composition of the CFGs from all sources. They all have a typical arabinoxylan structure with an Ara/Xyl ratio of about 0.6 except CFG-1 and 2 from WPEF which look more branched with 0.70 and 0.67 Ara/Xyl ratios, respectively. As reported in the literature they contain 31–36% Ara, 50–56% Xyl, 6–8% Gal, 1–6% Glc and 4–7% GlcA. Both NAG and MAG have a typical arabinogalactan composition with 31–46% Ara, 37–42% Gal, 2–12% Rha, 1–2% Xyl and 12–13% GlcA. The NAG is less branched with an Ara/Gal ratio of 0.75 in comparison to MAG in which the Ara/Gal ratio is 1.23.

3.4. Emulsifying properties

Beverage emulsions are a unique class of emulsions that differ from other food emulsions in that they are consumed in a highly diluted form. Since the beverage flavor emulsion is first prepared in a concentrated form and then diluted

Table 3
Carbohydrate composition of corn fiber and acacia gums (mole %)

Sugars	WPF		DPF		WPEF		Commercial gums		
	CFG-1	CFG-2	CFG-1	CFG-2	CFG-1	CFG-2	IG	NAG	MAG
Fuc	0.00	0.00	0.17	0.13	0.13	0.00	0.00	0.00	0.00
Rha	0.48	0.00	0.00	0.15	0.64	0.35	0.20	11.83	2.06
Ara	32.49	32.39	30.65	31.07	35.73	33.65	31.92	31.37	45.91
Gal	6.69	6.91	7.92	8.13	6.67	6.78	6.03	42.06	37.37
Glc	1.62	5.50	1.42	3.66	1.14	3.95	5.00	0.77	0.75
Xyl	54.09	50.90	55.75	51.46	50.99	50.21	50.16	0.64	2.30
GlcA	4.64	4.29	4.09	5.40	4.70	5.06	6.70	13.32	11.62
Total	100	100	100	100	100	100	100	100	100
Ara/Xyl ratio	0.60	0.64	0.55	0.60	0.70	0.67	0.64		
Ara/Gal ratio								0.75	1.23

Note: Fuc, Fucose; Rha, Rhamnose; Ara, Arabinose; Gal, Galactose; Glc, Glucose; Xyl, Xylose; GlcA, Glucuronic acid.

several hundred times with sugar solution to make the finished beverages, good stability is required in both the concentrated and diluted forms (Tan, 1998). In the soft drink, the most commonly used flavor oil is orange oil whose density is lower than water. To get the required ES, it has to be prepared properly in the presence of an emulsifier. The instability of an emulsion either in concentrated or diluted form is recognized as creaming (ringing), coalescence or flocculation. It is necessary to prepare emulsions with small droplets and narrow droplet size distribution to obtain a stable emulsion. To obtain small droplets, a two stage homogenization (Einhorn-Stoll et al., 2002; Elwell, Roberts, & Coupland, 2004) and high pressure treatments (Puppo et al., 2005) are recommended. Thus the coarse emulsion was prepared by using a high speed polytron homogenizer, which was further homogenized into a fine emulsion by passing through a high pressure EmulsiFlex-B3 homogenizer (see materials and method for detail). The emulsifying properties of corn fiber and acacia gums were evaluated by the photometric method of Pearce and Kinsella (1978), determining absorbancy at 650 nm and calculating the turbidity and emulsion stabilizing capacity. This method is a simple yet indirect study of emulsifying properties (Einhorn-Stoll et al., 2002; Buffo et al., 2001). The emulsion concentrates were diluted $125\times$ in the sugar solution and their absorbancy was measured immediately. Fig. 2A shows the turbidity of diluted emulsion immediately after its preparation (zero time). The droplet size in the well-emulsified solution was so homogeneous that at zero time every sample had similar activity. The turbidity of O-in-W emulsion without any emulsifying agent was as good as the turbidity of all the other O-in-W emulsions prepared in the presence of CFG or acacia gum emulsifiers, but the emulsions without emulsifier broke down in a few hours and oil droplets started floating at the top of the solution. But the emulsions prepared in the presence of the hydrocolloid (gum) did not reach that stage so quickly.

3.5. ES at room temperature

The ES can be defined as the maintenance of a homogeneous structure of the system. It was determined by measuring the absorbancy of the diluted emulsion from the clear droplet-depleted solution at the bottom of the bottles and converting into turbidity. The higher the turbidity means better ES. Figs. 2B–D, show the stabilities of O-in-W emulsion either in the absence or presence of emulsifiers after 1, 7 and 14 days, respectively. As expected, in the emulsions prepared without any emulsifier (W) oil droplets merge together to form a larger droplet. It leads to a decrease of the number of oil droplets and eventually causes the breakdown of the emulsion just in a day. Thus the turbidity of this emulsion dropped over $300\,\text{cm}^{-1}$ to about 50 just in one day and to about 10 and below 5 in 7 and 14 days, respectively. The solution starts getting clear from the bottom as its emulsion breaks and oil droplets float to the top. MAG, pretested by the supplier as a better emulsifier than NAG, showed similar ES until 1 day and superior after 7 days. The emulsion stabilities of CFG-1 and 2 from all three sources WPF, DPF and WPEF; and CFG (IG) look similar to each other after 1 day but superior to NAG and MAG. Their stabilities were better than NAG and superior or equal to MAG after 7 and 14 days. After 7 and 14 days, emulsion stabilities of both CFG-1 and 2 from WPEF and WPF were better than the CFG-1 and 2 from DPF. The CFGs with higher protein contents (Table 2) have better emulsion stabilizing capacities which correlates with the Randall, Phillips, and Williams (1988) hypothesis that gum arabic having high protein contents contribute more to their emulsifying capacities. The finding that the CFG-2 samples from all three CF sources have more protein than the CFG-1 samples from the respective sources and that they are better emulsifiers (Table 2, and Figs. 2 and 3) also suggests that the protein content of CFG samples may be partially or fully responsible for the emulsifying activity. This is the first documented evidence of this possibility.
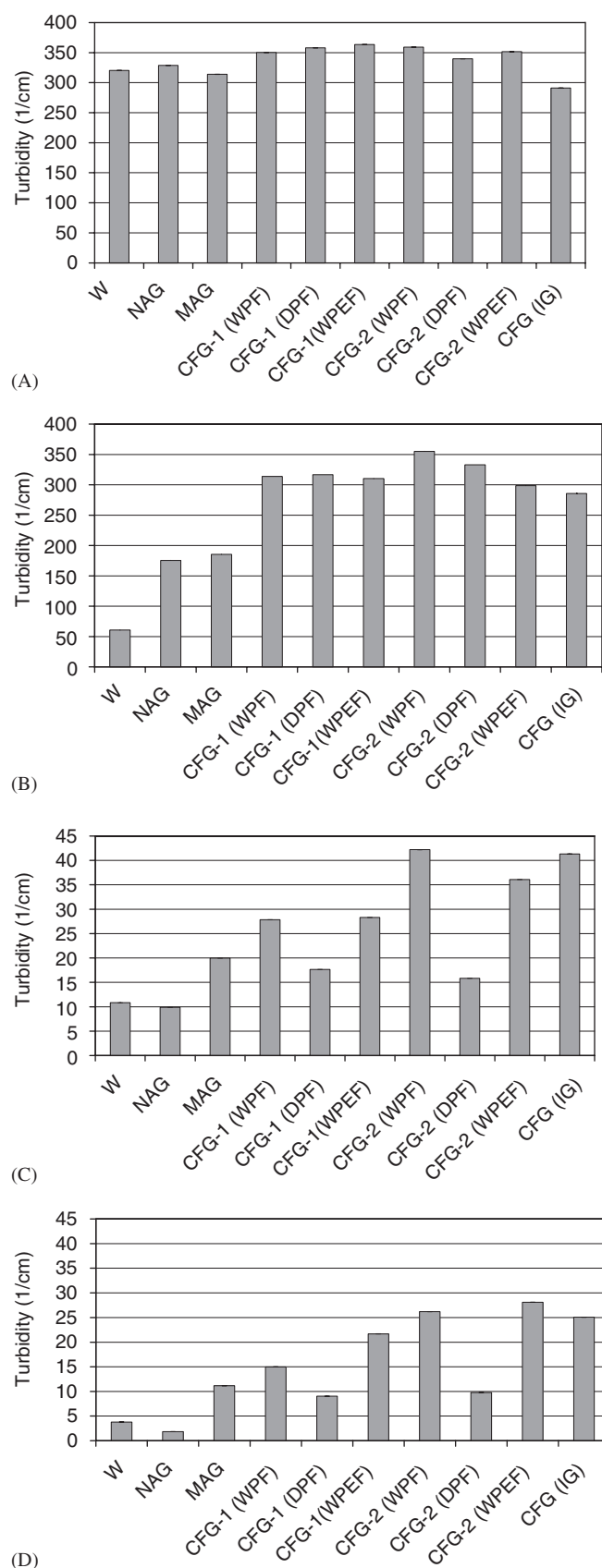
(A)

(B)

(C)

(D)

Fig. 2. Turbidity of gum samples at room temperature (A) right after preparing diluted emulsion and then after (B) 1 day (C) 7 days and (D) 14 days. Turbidity was measured by taking an aliquot from the bottom of the bottle. Higher turbidity indicates greater emulsion stability. Each plotted point is an average of three trials ±standard deviation.
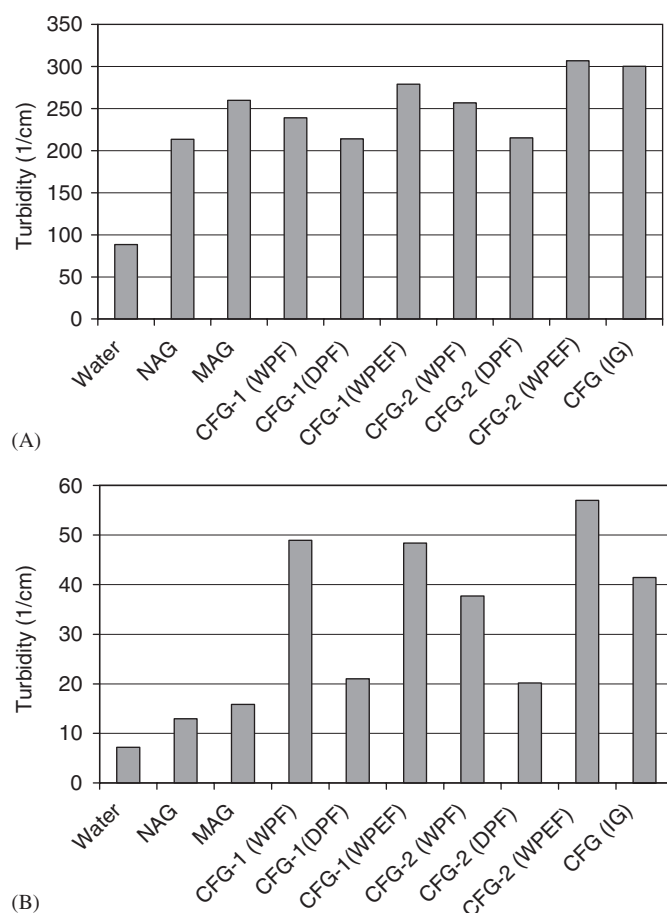

(A)

(B)

Fig. 3. Emulsion stability (turbidity) of gum samples at 45 °C after (A) 1 day (B) 7 days. Turbidity was measured by taking an aliquot from the bottom of the bottle. Higher turbidity indicates greater emulsion stability.
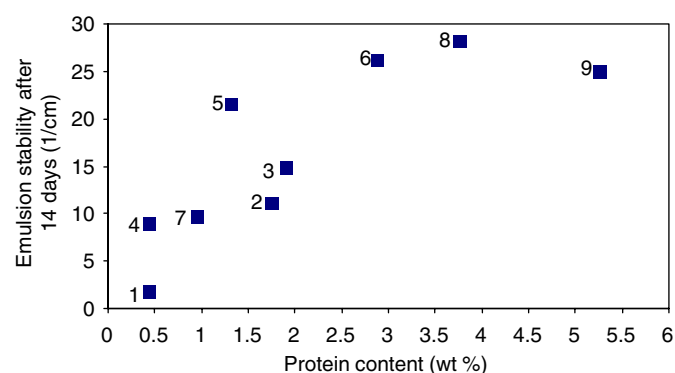

Fig. 4. Emulsion stability (turbidity) vs. protein content of all gum samples (see Fig. 2D for turbidity graph after 14 days and Table 2 for protein content), (1) NAG (2) MAG (3) CFG-1 (WPF) (4) CFG-1 (DPF) (5) CFG-1 (WPEF) (6) CFG-2 (WPF) (7) CFG-2 (DPF) (8) CFG-2 (WPEF) and (9) CFG (IG).

The correlation between the protein content and the ES after 14 days at room temperature for all samples is shown in Fig. 4. It is very interesting that the ES appears to correlate well with the protein content for the gum fractions isolated from the same source (e.g. CFG-2 forms more stable emulsion than the corresponding CFG-1).

But when the gum sample from one source is compared with the gum sample from different source, the ES does not correlate well with protein content (Fig. 4). Thus it can be interpreted that the protein is key to the emulsification process only in combination with some other structural and molecular characteristics of the gum sample. So it is essential to do the molecular characterization of these samples to make this interpretation clear and fully understand the molecular basis of their best emulsifying properties. The CFG-2 from DPF has more protein than the corresponding CFG-1, but its emulsion stabilizing capacity after 7 days both at room temperature and 45 °C looks slightly inferior to CFG-1 (Figs. 2C and 3B) but again looks superior after 14 days at room temperature (2D). The emulsifying property of these two gums might be very similar for 7 days and differ only after 14 days. The gums from DPF are the least effective emulsifiers among all CFGs and the difference in the emulsifying capacity of CFG-1 and 2 from this source is very minor. The emulsion stabilizing capacity of CFG-2 from WPEF among all the CFG samples is the maximum after 14 days at room temperature, which could be due to higher protein contents (Table 2) and/or a high level of branching in the polysaccharide backbone (Ara to Xyl ratio in Table 3). Gum arabic is described as a highly branched arabinogalactan bound to polypeptides that prevents aggregation by causing steric and electrostatic repulsion between oil droplets (Islam, Philips, Sljivo, Snowden, & Williams, 1997). CFG is also a highly branched polysaccharide that, at least in our preparations contains a tightly bound protein component. While little is known about the linkage, composition, or identity of the protein fraction associated with CFG, it is possible that CFG acts as an emulsifier by a similar mechanism as proposed for gum arabic.

3.6. ES at 45 °C

The purpose of these experiments was to examine the influence of temperature on the stability of emulsions prepared with gum samples. Figs. 3A and B show the ES after 1 and 7 days respectively at 45 °C for all the emulsions prepared in the presence or absence of gum emulsifiers. The ES of all the samples after 1 and 7 days at 45 °C are similar to their corresponding stabilities after 1 and 7 days at room temperature (Figs. 2B and C). At the higher temperature also CFGs from WPEF and WPF have greater emulsion stabilizing capacities than the CFGs from DPF.

4. Conclusions

CFGs extracted from WPF, DPF and WPEF of corn are similar in sugar composition but differ in protein content and sugar branching. CFGs extracted from WPEF and WPF have higher protein contents than those extracted from DPF. CFG-2 from each source has a higher protein content than the corresponding CFG-1. Both CFG-1 and 2 from WPEF are more branched (Ara/Xyl ratio in Table 3) than the other CFG samples. Emulsions prepared with CFGs from WPEF and WPF have higher stability than CFGs from DPF at both room temperature and 45 °C. CFG-2 is a better emulsifier than the corresponding CFG-1 from each source except CFG-2 from DPF which does not look superior to its corresponding CFG-1 after 7 days though it looks better after 14 days at room temperature. In general the CFG having higher protein content is generally a better emulsifier with the exception of the IG-CFG, which has the highest percent of protein (5.25%) in comparison to all CFG samples, but its emulsion stabilizing capacity is very close to that of CFGs from WPEF and WPF. This could be due to high ash content (16.97%) in this commercial, non-food grade CFG.

Emulsifying properties of CFG samples were generally superior or equal to the acacia gums in this experimental system (or in this application).

The high water solubility, low bulk viscosity, temperature stability (Whistler, 1993; Doner et al., 1998) and the high ability to stabilize oil droplets in the O-in-W emulsion system make CFG very promising for beverage emulsion stabilization.

Research to further study the molecular characteristics of CFG and the structure and linkage of its protein component is now underway. This knowledge may lead to a better understanding of CFG's emulsifying activity and present ways to enhance these effects.

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